

**Amendments to the Claims:**

This listing of claims will replace all prior versions, and listings, of claims in the application:

**Listing of Claims:**

1. (Currently Amended) For imaging a target object, an imaging system comprising:

a first beam splitter configured to substantially transmit part of received light as first transmitted light and to substantially reflect part of received light as first reflected light;

a defocus system configured to modify optical power of substantially one of the following: the first transmitted light and the first reflected light, and to transmit the same as first transmitted defocused light;

a reflector configured to reflect one of the following: the first reflected light and the first transmitted defocused light;

a second beam splitter configured to substantially transmit part of one of the following: the first transmitted light as second transmitted light and the first transmitted defocused light as second transmitted defocused light and configured to substantially reflect part of one of the following: the first transmitted defocused light as second reflected defocused light and the first reflected light as second reflected light; and

an imaging sub-system configured to focus one of the following pairs of light: the pair of the second transmitted light as imaged unaltered light and the second reflected defocused light, as imaged defocused light and the pair of the second transmitted defocused light as imaged defocused light and the second reflected light as imaged unaltered light and to focus the imaged defocused light with respect to an imaged defocused image plane and the imaged unaltered light with respect to an imaged unaltered image plane separated from the imaged defocused image plane, the second beam splitter oriented according to a mechanical angle such that the imaged unaltered light and the imaged defocused light have an angular separation other than zero.

2. (Currently Amended - Withdrawn) The system of claim 1 wherein the second beam splitter is further configured to transmit part of the first reflected light and ~~reflected~~ reflect part of the first transmitted light.

3 (Original) The system of claim 1 wherein the defocus system is a negative lens.

4 (Original) The system of claim 1, further comprising a collection system wherein the collection system is a lens.

5 (Withdrawn) The system of claim 1, further comprising an optical retardation plate and wherein the second beam splitter is a polarization beam splitter having a polarization beam splitter optical coating

6 (Original) The system of claim 1 wherein the imaging sub-system is an imaging lens

7 (Withdrawn) The system of claim 1 wherein the first beam splitter and the second beam splitter are polarization beam splitters having polarization beam splitter optical coatings.

8 (Currently Amended - Withdrawn) The system of claim ~~1~~12, further comprising a second imaging sub-system and a second detector.

9. (Currently Amended) The system of claim ~~1~~12 wherein the detector has first and second focus areas.

10. (Withdrawn) The system of claim 1, further comprising a spectral dispersing element configured to transmit light to the imaging sub-system.

11. (Currently Amended - Withdrawn) The system of claim 1, further comprising:

a secondly-oriented first beam splitter configured to substantially transmit part of the collected light as first transmitted light and to substantially reflect part of the collected light as first reflected light;

a secondly-oriented defocus system configured to modify optical power of substantially one of the following: the first transmitted light and the first reflected light to transmit as defocused light;

a secondly-oriented reflector configured to reflect one of the following: the first reflected light and the defocused light;

a secondly-oriented second beam splitter configured to substantially transmit part of one of the following: the first transmitted light as second transmitted first transmitted light and the defocused light as second transmitted defocused light and configured to substantially reflect one of the following: the defocused light as second reflected defocused light and the first reflected light as second reflected first reflected light;

a secondly-oriented imaging sub-system configured to focus one of the following pairs of light: the pair of the second transmitted first transmitted light as imaged unaltered light and the second reflected defocused light as imaged defocused light and the pair of the second transmitted defocused light as imaged defocused light and the second reflected first reflected light as imaged unaltered light and to focus the imaged defocused light with respect to a imaged defocused image plane and the imaged unaltered light with respect to an imaged unaltered plane being separated from the imaged defocused plane; and

a secondly-oriented first detector positioned to receive the imaged defocused light and the imaged unaltered light, the secondly-oriented second beam splitter oriented according to a mechanical angle such the unaltered light and the defocused light have an angular separation.[[.]]

12. (Currently Amended) The system of claim 1, further including a first detector positioned to receive the imaged defocused light with respect to the imaged defocused image plane and the imaged unaltered light with respect to the imaged unaltered image plane.

13. (Original) For imaging a target object, an imaging system comprising:  
a first beam splitter configured to substantially transmit part of collected light as first transmitted light and to substantially reflect part of collected light as first reflected light;  
a defocus system configured to modify optical power of substantially one of the following: the first transmitted light and the first reflected light to transmit as defocused light;  
a reflector configured to reflect one of the following: the first reflected light and the defocused light;  
a second beam splitter configured to substantially transmit part of one of the following: the first transmitted light as unaltered light and the defocused light as defocused light and configured to substantially reflect part one of the following: the defocused light as defocused light and the first reflected light as unaltered light; and  
a first detector positioned to receive the defocused light and the unaltered light with respect to a first object plane and the defocused light with respect to a second object plane, the defocus system configured to modify optical power such that the first depth of focus overlaps the second depth of focus, the second beam splitter oriented according to a mechanical angle such the unaltered light and the defocused light have an angular separation.

14. (Original) For imaging a target object, an imaging system comprising:  
a first beam splitter configured to substantially transmit part of collected light as first transmitted light and to substantially reflect part of collected light as first reflected light;  
a defocus system configured to modify optical power of substantially one of the following: the first transmitted light and the first reflected light to transmit as defocused light;  
a reflector configured to reflect one of the following: the first reflected light and the defocused light; and

a second beam splitter configured to substantially transmit part of one of the following: the first transmitted light as unaltered light and the defocused light as defocused light and configured to substantially reflect part one of the following: the defocused light as defocused light and the first reflected light as unaltered light, the second beam splitter oriented according to a mechanical angle such the unaltered light and the defocused light have an angular separation.

15. (Original) For imaging a target object, a method comprising:

substantially transmitting part of received light as first transmitted light and to substantially reflect part of received light as first reflected light;

modifying optical power of substantially one of the following: the first transmitted light and the first reflected light, and to transmit the same as first transmitted defocused light;

reflecting one of the following: the first reflected light and the first transmitted defocused light;

transmitting part of one of the following: the first transmitted light as second transmitted light and the first transmitted defocused light as second transmitted defocused light and configured to substantially reflect part of one of the following: the first transmitted defocused light as second reflected defocused light and the first reflected light as second reflected light; and

focusing one of the following pairs of light: the pair of the second transmitted light as imaged unaltered light and the second reflected defocused light, as imaged defocused light and the pair of the second transmitted defocused light as imaged defocused light and the second reflected light as imaged unaltered light and to focus the imaged defocused light with respect to an imaged defocused image plane and the imaged unaltered light with respect to an imaged unaltered image plane separated from the imaged defocused image plane, the second beam splitter oriented according to a mechanical angle such the imaged unaltered light and the imaged defocused light have an angular separation other than zero.

16. (Withdrawn) For imaging a target object, an imaging system comprising:

- a first imaging sub-system comprising a light collection system positioned with respect to the target object in a first orientation, the first imaging sub-system configured to transmit a first image of the target object;
- a first detector positioned to receive the first image of the target object at a first location on the first detector when the target object is in a first target location and to receive the first image of the target object at a second location on the first detector when the target object is in a second location different than the first location of the target object;
- a second imaging sub-system comprising a light collection system positioned with respect to the target object in a second orientation different than the first orientation, the second imaging sub-system configured to transmit an second image of the target object at a best focus distance from the second imaging sub-system;
- a second detector positioned to receive the second image of the target object; and
- a processor communicatively linked to the first detector and configured to determine a distance between the first location on the first detector and the second location on the first detector, the processor communicatively linked to the second imaging sub-system and configured to transmit instructions to the second imaging sub-system based upon the determined distance between the first location on the first detector and the second location on the first detector, the second imaging sub-system configured to change the best focus distance from the second imaging sub-system based upon the instructions received from the processor to correct focus.

17. (Withdrawn) The system of claim 16 wherein the first orientation is along an x-axis and the second orientation is along a y-axis, the x-axis and y-axis being substantially perpendicular to one another.

18. (Withdrawn) For imaging a target object in a flow stream, an imaging system comprising:

a first imaging sub-system comprising a light collection system positioned with respect to the target object in a first orientation, the first imaging sub-system configured to transmit a first image of the target object;

a first detector positioned to receive the first image of the target object at a first location on the first detector when the target object is in a first target location and to receive the first image of the target object at a second location on the first detector when the target object is in a second location;

a second imaging sub-system comprising a light collection system positioned with respect to the target object in a second orientation, the second imaging sub-system configured to transmit an second image of the target object;

a second detector positioned a distance along an optical axis from the second imaging sub-system to receive light from the second imaging sub-system; and

a processor communicatively linked to the first detector, the processor configured to determine a distance between the first location on the first detector and the second location on the first detector, the processor communicatively linked to the second imaging sub-system, the processor configured to transmit instructions to the second imaging sub-system based upon the determined distance between the first location on the first detector and the second location on the first detector, the second imaging sub-system configured to change the distance that the second detector is positioned along the optical axis from the second imaging sub-system based upon the instructions received from the processor.

19. (Withdrawn) The system of claim 18 wherein the first imaging sub-system has a magnification and the instructions are based on the determined distance multiplied by the square of the magnification of the second imaging sub-system.

20. (Withdrawn) A method for maintaining focus in an imaging system comprising:

segmenting objects of interest;

analyzing frequency content of one or more imaged objects in two focus areas associated with received unaltered and defocused light;

if frequency content is not balanced, calculating a focal error signal;

if frequency content is not balanced, determining focal shift; and

adjusting refocusing optics based upon the determined focal shift and the focal error signal.

21 (Withdrawn) The method of claim 20 wherein the frequency content of the one or more imaged objects in the two focus areas is balanced if the frequency content in the two focus areas is equal.

22. (Withdrawn) The method of claim 20 wherein the unaltered and defocused light is received on one detector.